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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Group: 1638

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Confirmation No.: 1633

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Application No.: 10/664,658

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Invention: METHODS AND COMPOSITIONS FOR
ENHANCED PLANT CELL
TRANSFORMATION

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Applicant: Stanton B. Gelvin et al.

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Filed: September 18, 2003

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*Attorney
Docket:* 3220-94790

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Examiner: ZHENG, L.I.

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DECLARATION UNDER 37 C.F.R. §1.132

Commissioner of Patents
P.O. Box 1450
Alexandria, VA 22313-1450

1. I, Stanton B. Gelvin, am a co-inventor of the patent application referenced above.
2. I have read the Office Action mailed December 19, 2006 and reviewed the application as filed and the pending claims.
3. The application as filed describes methods by which it was shown that overexpression of H2A leads to increased transformation (see for example, page 9, first and second paragraphs; page 10, third paragraph; and page 11, first paragraph) and describes "histone genes" or "a histone gene" generally, e.g. page 3, fifth paragraph, page 4, first paragraph, and original claims.
4. The same methods used to establish the utility of overexpression of *H2A* in increasing

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cell transformation have now been applied to other histones to demonstrate that overexpression of histone genes other than *H2A* in plants, increase transformation efficiency to about two-fold as compared to the wild-type control. For example, histone genes representing core histone groups *H3R* and *H4O*, upon overexpression, show increased transformation efficiency as compared to the wild-type control. A summary of the results obtained is provided in attached Exhibit A.

4. Using similar vectors, constructs, and methods as taught in the specification, co-electroporation of tobacco BY-2 protoplasts with the *H2A1* gene resulted in a three-fold increase in transformation efficiency as compared to an empty vector. Summary of the results are presented in the attached Exhibit B.

5. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the Untied States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully Submitted,

Dated: 6/8/08


Stanton B. Gelvin

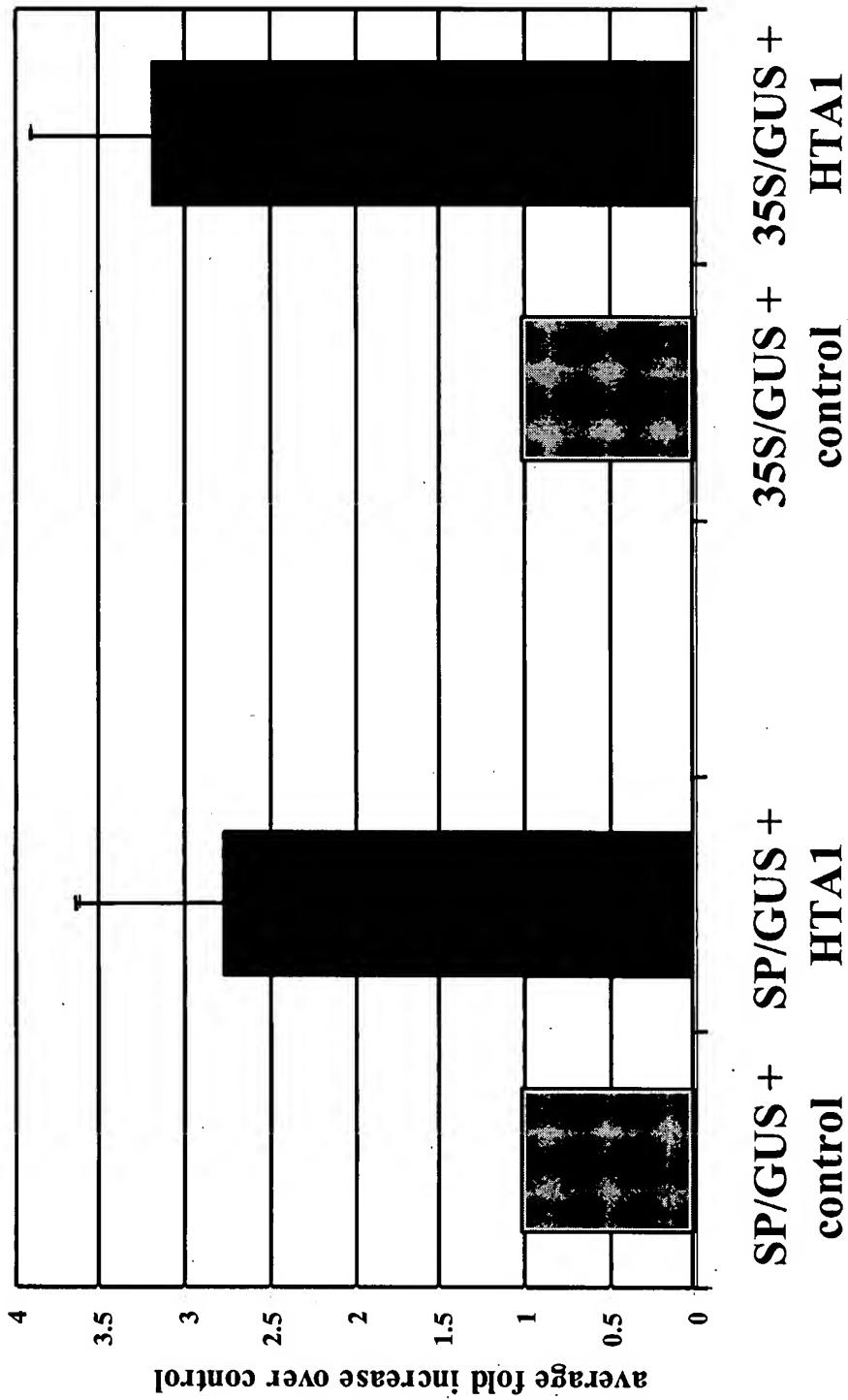
EXHIBIT A

Primary screening to determine which core histone genes, when over-expressed, can enhance *Agrobacterium*-mediated transformation of *Arabidopsis*

Core Histone Gene	Total # of transformed plants screened	Individual line ID number with increased response to <i>Agrobacterium</i> -mediated transformation	
		Klett 0.1	Klett 0.01
HFO3	20	3; 4; 9; 11; 15; 19	3; 5; 15; 18; 19
HTA1	20	4; 6; 14; 16	6; 9; 14
HTA2	14	5; 11; 13	4; 5; 10; 11; 14
HTA3	18	2; 4; 5; 6; 7; 10; 11; 13; 14; 15; 19	1; 4; 5; 7; 9; 10; 11; 13; 14; 15; 17
HTA5	18	1; 5; 6; 11; 15; 16; 18	-
HTA6	5	5	1
HTA6	20	2; 4; 7; 9; 14; 17; 20	6; 9; 17; 20
HTA8	20	-	-
HTA10	-	-	-
HTB1	18	10; 12; 18	5; 8; 10; 15
HTB1	18	5; 8; 10; 15	8; 10; 12; 18
HTB3	6	-	-
HTB5	20	9	-
HTB8	20	6; 18; 20	1; 3; 7; 9
HTB9	9	9	5
HTB10	20	8	8
HTB11	20	13	15
HTR4	20	5; 10; 12	3; 4; 13; 17
HTR11	20	1; 2; 3; 4; 5; 7; 8; 10	7; 8; 16; 19; 20
HTR13	20	6; 11; 13; 16; 17	-

A total of 22 representative core histone genes were tested by expressing cDNAs of these genes under the CaMV 35S promoter in transgenic *Arabidopsis* plants. Infections were performed at a bacterial concentration of 10^5 cells/ml (Klett=0.01) or at 10^6 cells/ml (Klett=0.1).

EXHIBIT B



Numbers represent the percentage of GUS positive protoplasts vs. control for three independent electroporation experiments.

More than 10,000 cells were examined for each treatment.